

FIGURE 1

Pvs28 Nucleotide Sequence

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40      tccactctctctgttccacacittatctttgtttcccc
      ccattcggccaccacacigcattatatacaaaaacgactcccc
80      clttgagataaacaccccaacigagctcgtattccccctcccc
      acttttgcgcccctctgttcaaaatgaataacctacca
120      cagcttgcctgttccctctggccatcgtgcttactgttaag
      cacaccttcgcaaaaggcaccgaggagaccaatgcaaaa
200      atggctatgtagtccaaatgagcaatcatcttggaaatgcaa
      atgcaaacgacgggtttgtttatggcaaatgaaaacacttgc
280      gaggaaaaaacgctgattgcacaaatccacaaaaatgtaaal
      aaaacigtggagactacgctgtgtgtgtgcaaacaccagaaat
360      gaaatgtagtggaaaggagcattacgagtcggcctgcatatta
      gggtacaccgtaaatgaaatgagggtgtgtacttccaataaat
440      gtaacggcgttttgtgtgtgaaaggaaaaagtgcatactaga
      tcccgctaatgtgaaacagcaccatgtgtcttctgttaatala
520      ggtaaccacatlggtatgaaatcaaaaaatgttggaagaaggccag
      gaaaaaacgtgaaatgcacgttgaaggtgtaaggcaaacgaaaga
600      atgtaaaggagacacagaattattacaaggctgtgtgtggaag
      ggaaagcggcgggaaggagcaggcgtgtgtggaagaaggcaggc
680      gaaaggcggcggcgggaaggagcaggcgtgtgtggaagaaggc
      cgggtgggaagacacagggagcaggcttacagctctcaltgaacggga
760      tctgcagttaatacagcatactactgttatttgccttcttca
      tgaatgattagtgtagacgattctacacacacacacacaaa
840      catacacaagggtgggaaggcgtctcacagagtcagttcaagg
      tcatacgcacaaaaaaaggaaaagtacatccagcctgggtgaaa
920      gagcatltagtgtgtcagttatcttgggtggaaggcacccct
      ccacccagttgcgtgtgtgttaccttaaaacttagtggcca
1000
1040      cccatatacgaatttgactttgtctcgc
1066

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FIGURE 2**Pvs28 Amino Acid Sequence**

N-terminal-

MNTYHSLLELLAIVLTVKHTEFAKVTAETQCKNGYVVQMSNHFECKCNDGFVMANENTCEEKRDCTNPQNVNKNCGDYAVCA
NTRMNDEERALRCGCILGYTVMNEVCTPNKCNGVLCGKGKCILDPANVNSTMCSNIGTTLDESKCGKPGKTECTLKCKANE
ECKETQNYKCVAKGSGGEGSGGEGSGGGGGTGAAYSLMNGSAVISILLVFAFFMMSLV

-C-terminal

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FIGURE 3**Pvs25 Nucleotide Sequence**

5'- CTGACTTTCGTTTCACAGCACTGATTTTTTTGTTTCGACCGCTCAATTTCGC
CACTTGCCATTTCGATTGTTTGCTTGTGCTTTTGTCTTATTCGCCC
GTTTTCCGCTTGCCCGTTCGCCCGCTCCACAACGGCGCGTGCAAAGGT
TGCCCAACCACCGACCACAAAACCTTATTCACCACCATCCGAGCGGAAAGG
AAGCGGCCACTGTGCTGCTACCTCCCCGAATAACAACCTCCACTTAGC
CAAAATGAACCTCTACTACAGCCTCTTCGTTTTTTTCTCTGTCCTCAAAATTG
CGCTAAAGTATAGCAAGGACGCCGTACGGTAGACACCATATGCAAAAAT
GGACAGCTGGTTCAAAATGAGTAACCACTTTAAAGTGTATGTGTAAACGAAGG
GCTGTGCACCTTCCGAAAAATACATGTGAAGAAAAAAATGAATGCAAGA
AAGAAACCCCTAGGCAAGCATGCGGGGAATTTGGCCAGTGTATAGAAAAC
CCAGACCCAGCACAGGTAAACATGTACAAATGTGGTTGCATTGAGGGCTA
CACTTTGAAGGAAGACACTTGTGCTTGATGTATGTCAATACAAAAATT
GTGGAGAAAAGTGGCGAATGCATTGTTGAGTACCTCTCGGAAATCCAAAGT
GCAGGTGCTCATGTGCTATTGGCAAAAGTCCCCAATCCAGAAAGATGAGAA
AAAATGTACCAAAACGGGAGAACTGCTTGTCAATTGAAATGTAAACACAG
ATAATGAAGTCTGCAAAAATGTTGAAGGAGTTTACAAGTCCAGTGTATG
GAAGGCTTTACGTTTCGACAAAGAGAAAAATGTATGCCTTTCCTATTCTGT
ATTTAACATCCTAACTACTCCCTCTCTTTATCATCCTGCTGTGTCCTT
CGTACGTCATATAAGTGCAGAACTTGCGCAGCTAAGCAGCGCAAAATTTT
TAAGTTAAAAATACTTTTCTTTACTGAACCTTACCGACTTGTGATGT
-3'

FIGURE 4**Pvs25 Amino Acid Sequence**

N-terminal-
MNSYYSLFVFFLVQIALKYSKAAVTVDTICKNGQLVQMSNHFKCMCNEGL
VHLSNTCEEKNECKKETLGKACGEFGQCIENPDPAQVNNMYKCGCIEGYT
LKEDTCVLDVCQYKNCGESGECIVEYLSEIQSAGCSAIGKVPNPEDEKK
CTKTGETACQLKCNVDNEVCKNVEGVYKQCQCMEGFTFDKEKNVCLSYSVF
NILNYSLFFIILLVLSYVI
-C-terminal

FIGURE 5

Pvs25-Pvs28 Fusion Protein Amino Acid Sequence

N-terminal-

AVTVDTICKNGQLVQMSNHFKCMCNEGLVHLSSENTCEEKNECKKETLGKACGEFGQCIENTPDPAQVNMVYKCGCIEGYTLKED
TCVLDVCQYKNCGESGECIVEYLSEIQSAGCSAIGKVPNPEDEKKCTKTGETACQLKCNSTDNEVCKNVEGVYKQCQCMEGFTF
DKEKNVCLS GGGPGGG AKVTAETQCKNGYVQMSNHFECKCNDGFVMANENTCEEKRDCTNPQNVNKNCGDYAVCANT
RMNDEERALRCGILGYTVMNEVCTPNKCNGLCGKGKCLDPANVNSTMCSNIGTTLDESKKCGKPGKTECTLKCKANEEC
KETQNYKCVAKSGGEGSGGEGSGGEGSGGGTGAAYSLMN

-C-terminal

GGGPGGG linker sequence underlined.

Figure 6

EAEASAVTDTICKNQLVQMSHHFKMCNEGLVHLSENTECKNECKKET 50
LGKACGEFGQCIENPDPAQVMYKKGCIEGYTLKEDTCVLDCQYKNCGES 100
GECIVEYLSEIQSAGSCSAIGKVPPEDEKKCTKTGETACQLKCNTDNEVC 150
KNVEGVYKQCQCMEGFTFCREKNVCLGPHHHHHH 186 (SEQ ID NO:16)

EAEASKVTAETQCKNGYVVMQSNHFECKNDGFVLANENTCEERDCTNP 50
 QNVNKNCGDYAVCANTRMNEERALRCGILGYVMNEVCTPYKNGVLC 100
 GKKGKCLDPANVNSTMCSNIGSTLDESKCKPGKTECTLKCRANECK 150
 ETQNYVKCAKSGGEGSGGEGSGGEGSGGEGSGGEGGGDTGAAYSGPH 200
 HHHHH 205 (SEQ ID NO:17)

EAEASKVTAETQCKNGYVVQSMNHFECKNDGFLANENTCEEKRDCTNP 50
 QNVNKNCGDYAVCANTRMNEERALRCGILGYTVNNEVCTPYKNGVLC 100
 GKGCILDPAVQSTMCNIGSTLDESKKCGPKTECTLCKRANECK 150
 ETQNYKCVAKSGGEGSGGEGSGGEGSGGEGSGGEGSGGDTGAAYSGPH 200

HHHHH 205 (SEQ ID NO: 18)

EAEASKVTAETQCKNGYVVMQSNHFECKNDGFLVANENTCEEKRDCTNP 50
QNVNKNCGDYAVCANTRMNEERALPCGCILGYVTVMNEVCTPYKCNGLVC 100
GKGKCILDPAVNSTMCSCNIGSTLDFSKKCGKPGKTECTLCKKANECK 150
ETQNYKCVAKGPHHHH 169 (SEED ID NO:19)

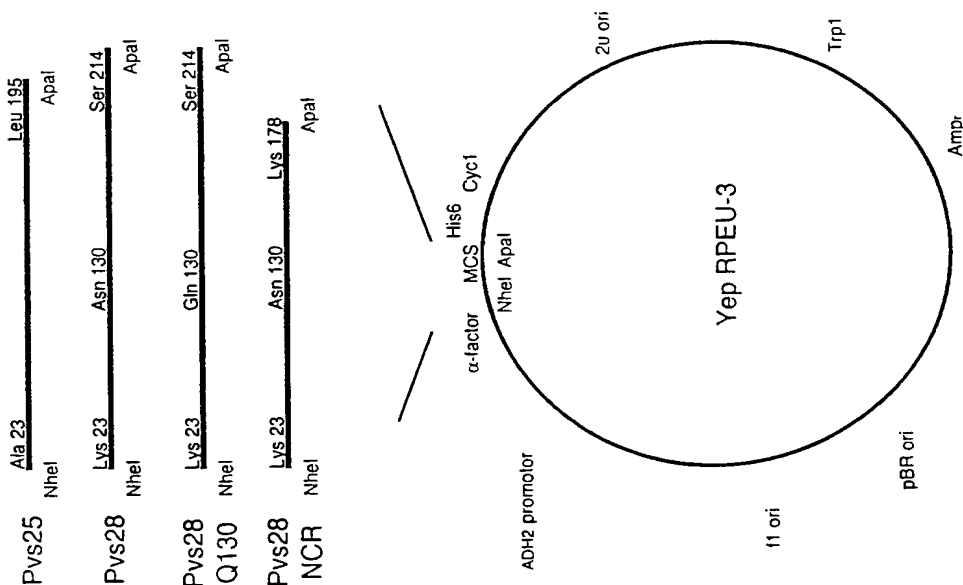
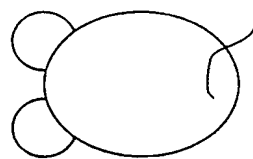
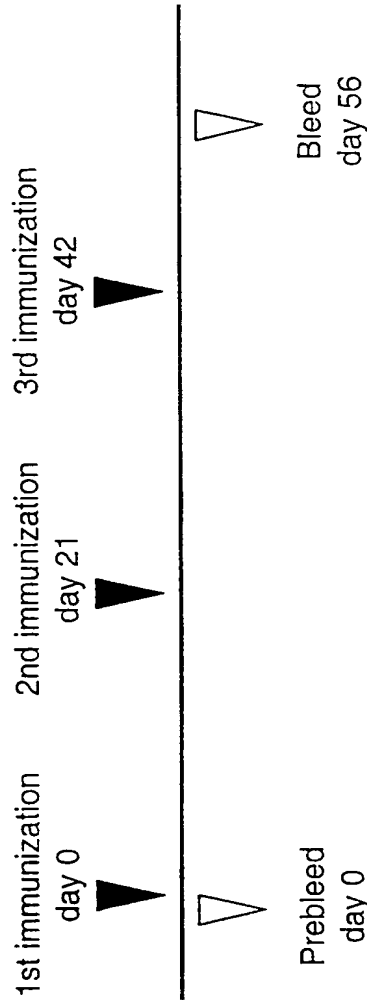


Figure 7

Experimental protocol for immunization with Pvs25 and Pvs28



Female, 8wks old
BALB/c (H-2d)
C57BL/6 (H-2b)
A/J (H-2a)
B10.BR (H-2k)
CAF1 (H-2a/d)



<u>Immunization</u>	<u>Groups</u>
Intraperitoneal injection with 50 µg of protein absorbed by 800 µg of Alum in 500 µl of PBS pH7.2	1 Alum alone 2 Pvs25 /Alum 3 Pvs28 /Alum